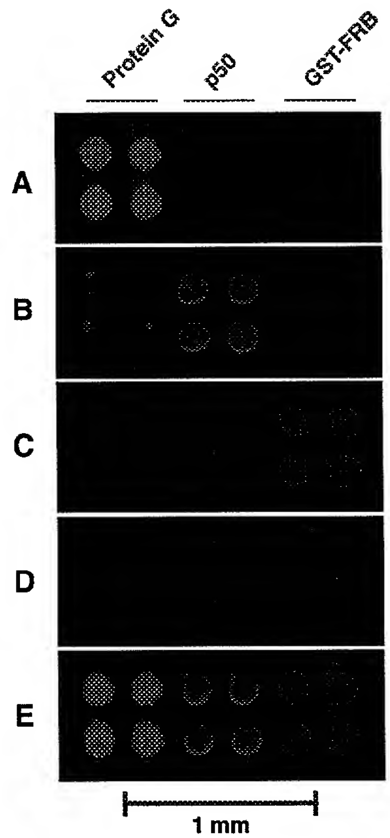

Fig. 1. Detecting protein-protein interactions on glass slides. (**A**) Slide probed with 0.5 ug/mL BODIPY-FL-IgG. (**B**) Slide probed with 0.1 ug/mL Cy3-IKBa. (**C**) Slide probed with 0.5 ug/mL Cy5-FKBP12 + 100 nM rapamycin. (**D**) Slide probed with 0.5 ug/mL Cy5-FKBP12 (no rapamycin). (**E**) Slide probed with 0.5 ug/mL BODIPY-FL-IgG + 0.1 ug/mL Cy3- IKBa + 0.5 ug/mL Cy5-FKBP12 + 100 nM rapamycin. In all panels, BODIPY-FL, Cy3, and Cy5 fluorescence were false-colored blue, green, and red, respectively.

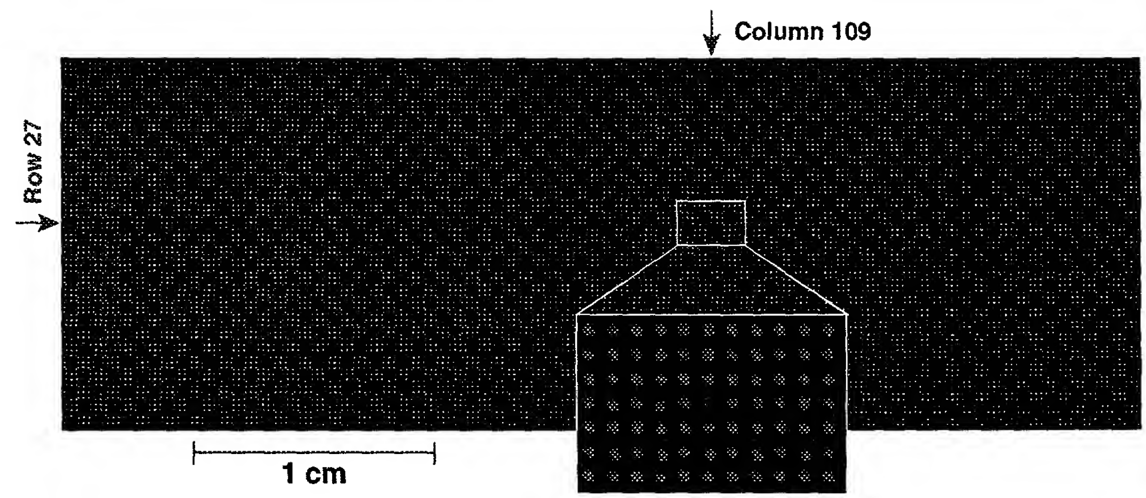

Fig. 2. 10,800 spots on a single slide. Protein G was printed 10,799 times. A single spot of GST-FRB was printed in row 27, column 109. The slide was probed with 0.5 ug/mL BODIPY-FL-IgG + 0.5 ug/mL Cy5-FKBP12 + 100 nM rapamycin. BODIPY-FL and Cy5 fluorescence were false-colored blue and red, respectively.

http://www.jbc.org/ by guest on June 14, 2019

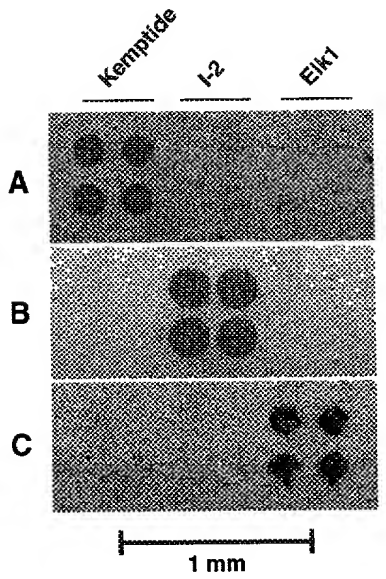

Fig. 3. Detecting the substrates of protein kinases on glass slides. (**A**) Slide incubated with the catalytic subunit of cAMP-dependent protein kinase (PKA). (**B**) Slide incubated with casein kinase II (CKII). (**C**) Slide incubated with p42 MAP kinase (Erk1).

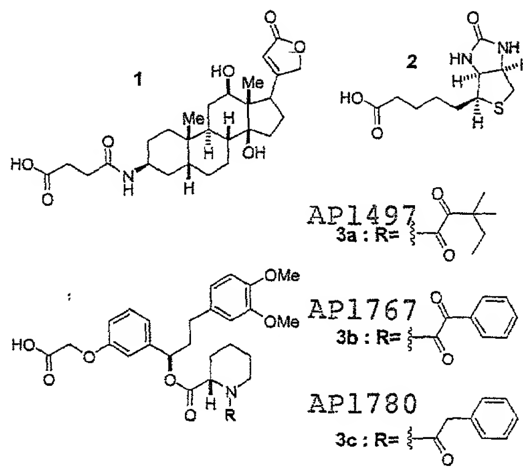

Fig. 4. Compounds used for the identification of the targets of small molecules. All compounds were coupled to bovine serum albumin through their carboxylate groups (either directly or via a flexible linker).

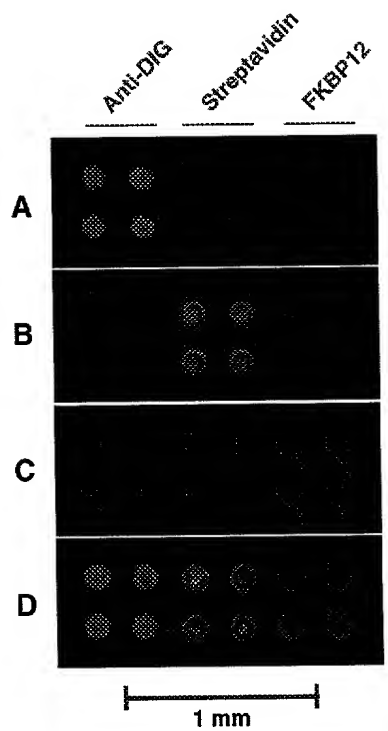

Fig. 5. Detecting the targets of small molecules on glass slides. (**A**) Slide probed with 10 ug/mL Alexa488-BSA-**1**. (**B**) Slide probed with 10 ug/mL Cy5-BSA-**2**. (**C**) Slide probed with 10 ug/mL Cy3-BSA-**3a**. (**D**) Slide probed with 10 ug/mL Alexa488-BSA-**1** + 10 ug/mL Cy5-BSA-**2** + 10 ug/mL Cy3-BSA-**3a**. In all panels, BODIPY-FL, Cy3, and Cy5 fluorescence were false-colored blue, green, and red, respectively.

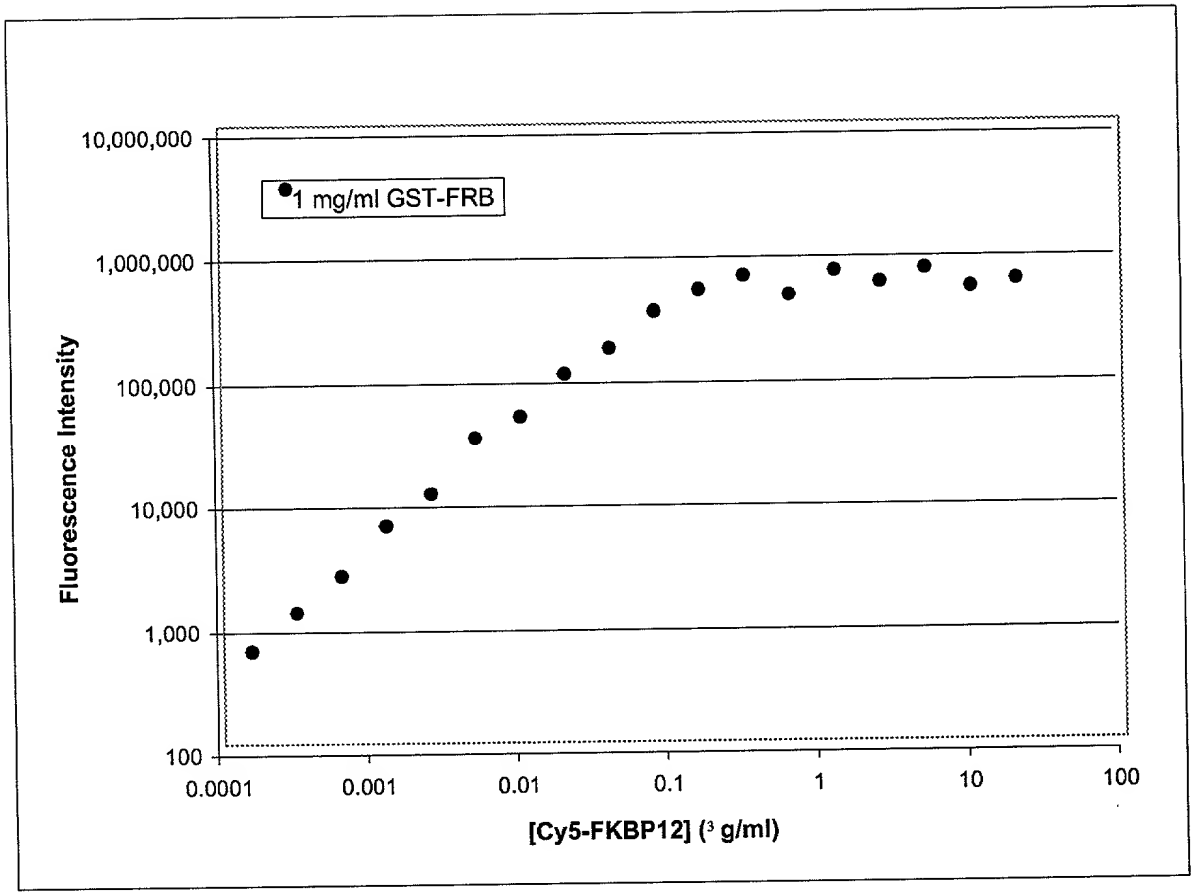

Fig. 6. Fluorescence intensity scales linearly with the concentration of solution-phase protein over four orders of magnitude. FRB was spotted on aldehyde slides in triplicate at a concentration of 1 mg/ml. The slides were then probed with Cy5-FKBP12, ranging in concentration from 150 pg/ml to 20 ug/ml. All solutions contained 1 uM rapamycin.

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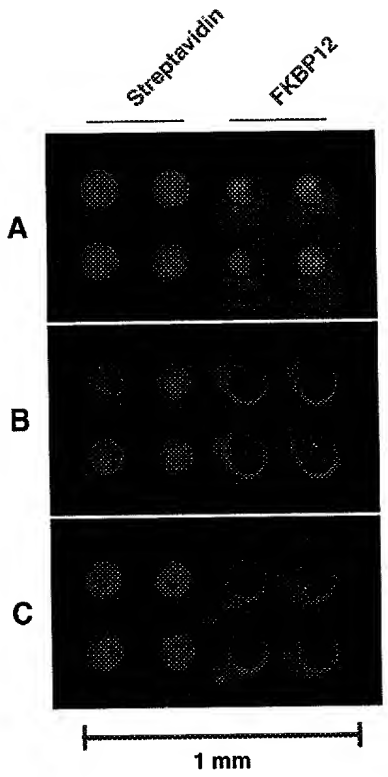
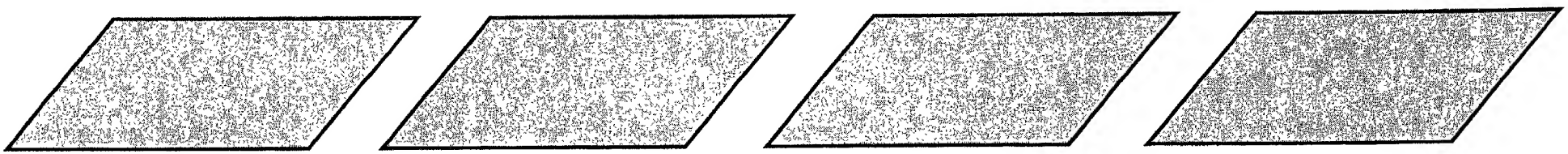


Fig. 7. Detecting the targets of low-affinity ligands on glass slides. (**A**) Slide probed with Cy5-BSA-**2** + Cy3-BSA-**3a**. (**B**) Slide probed with Cy5-BSA-**2** + Cy3-BSA-**3b**. (**C**) Slide probed with Cy5-BSA-**2** + Cy3-BSA-**3c**. All conjugates were used at a concentration of 10 ug/ml. In all panels, Cy3 and Cy5 fluorescence were false-colored green and red, respectively.

SCREENING METHOD 1



aldehyde or BSA-NHS slides

FIGURE 8A

TOE080" EH2E2660

SCREENING METHOD 1

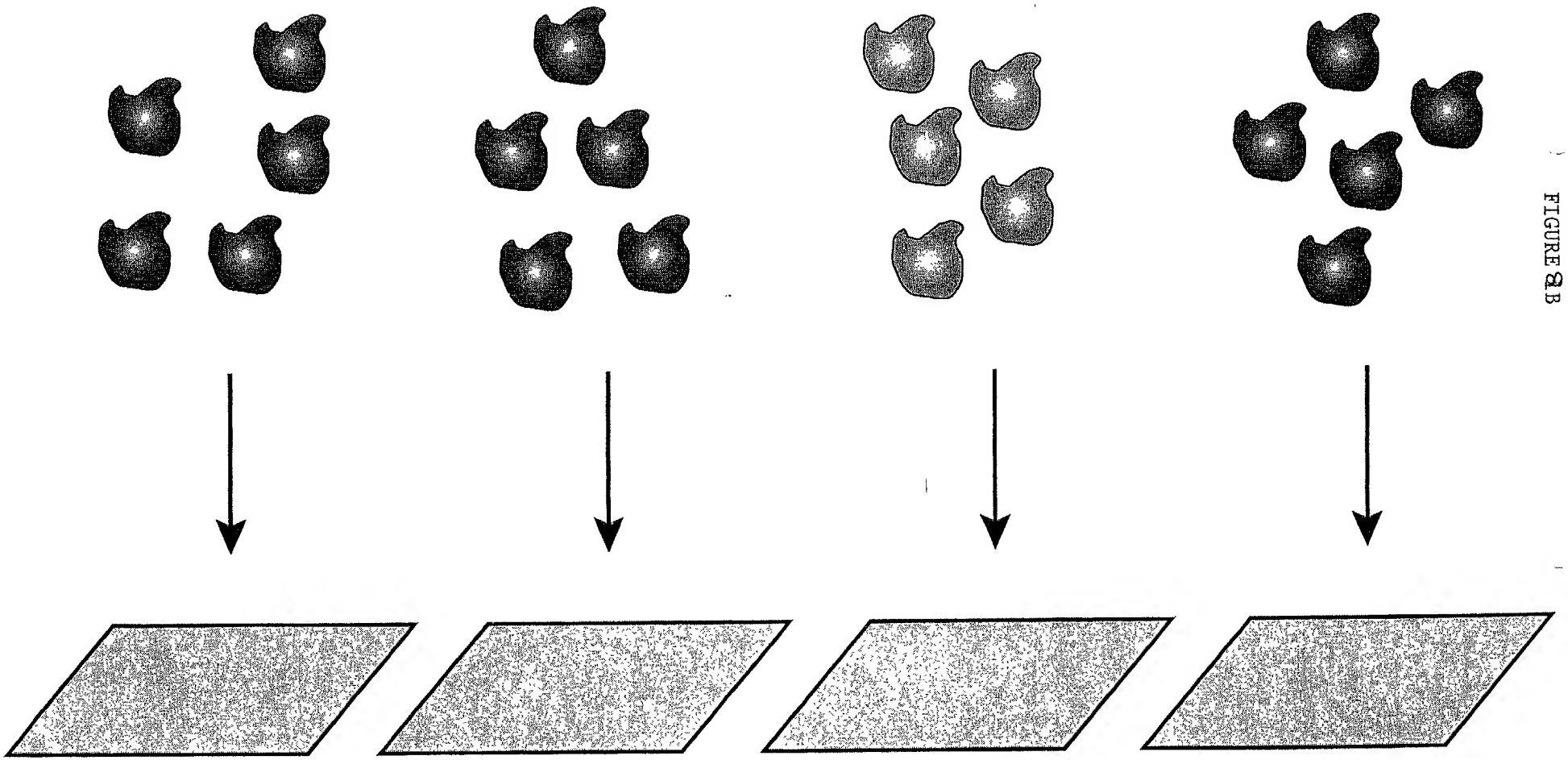


FIGURE 8A B

TOE080" E+2E2660

SCREENING METHOD 1

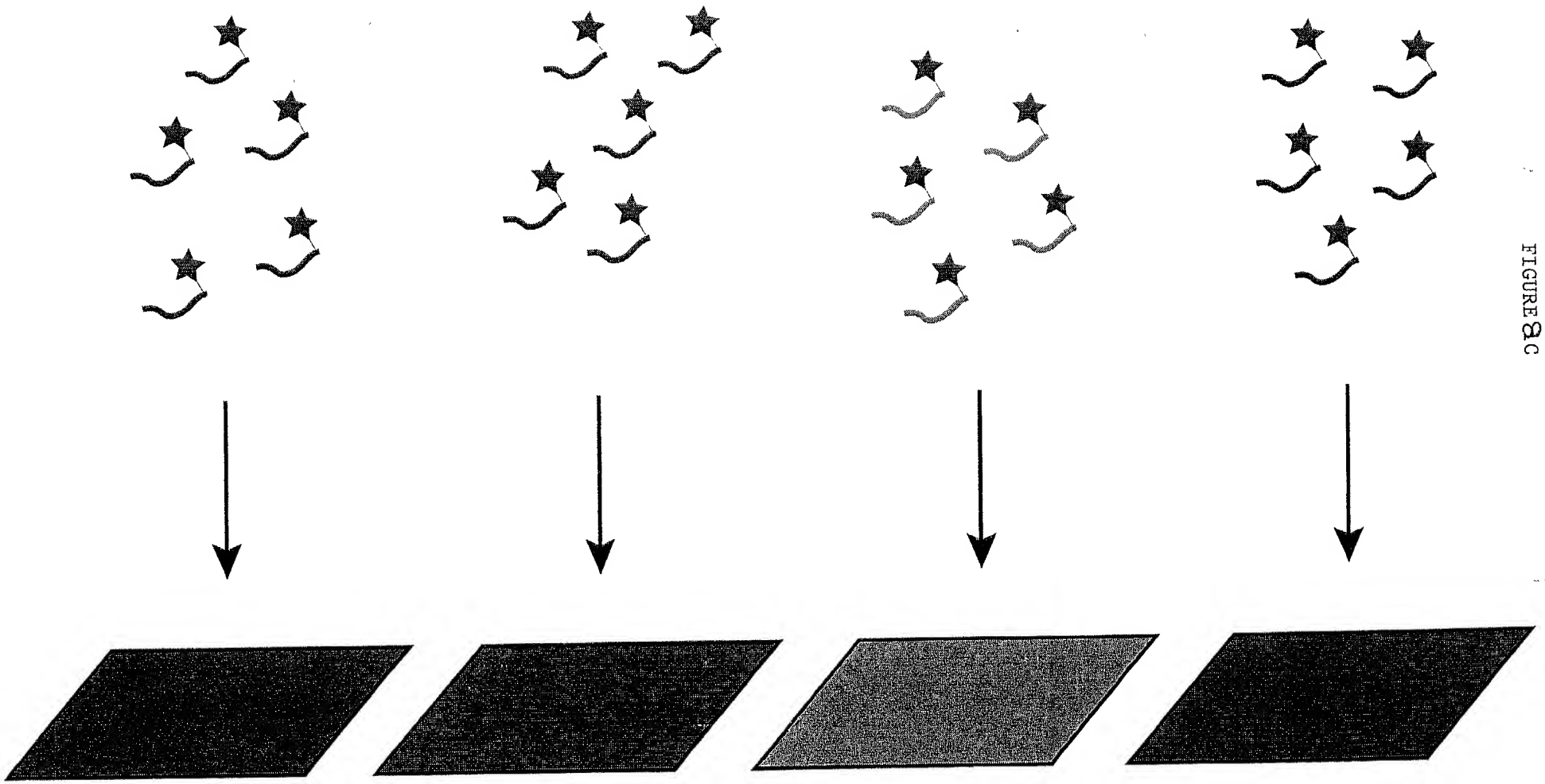


FIGURE 8C

TOED80 "Eh2E2660

SCREENING METHOD 1

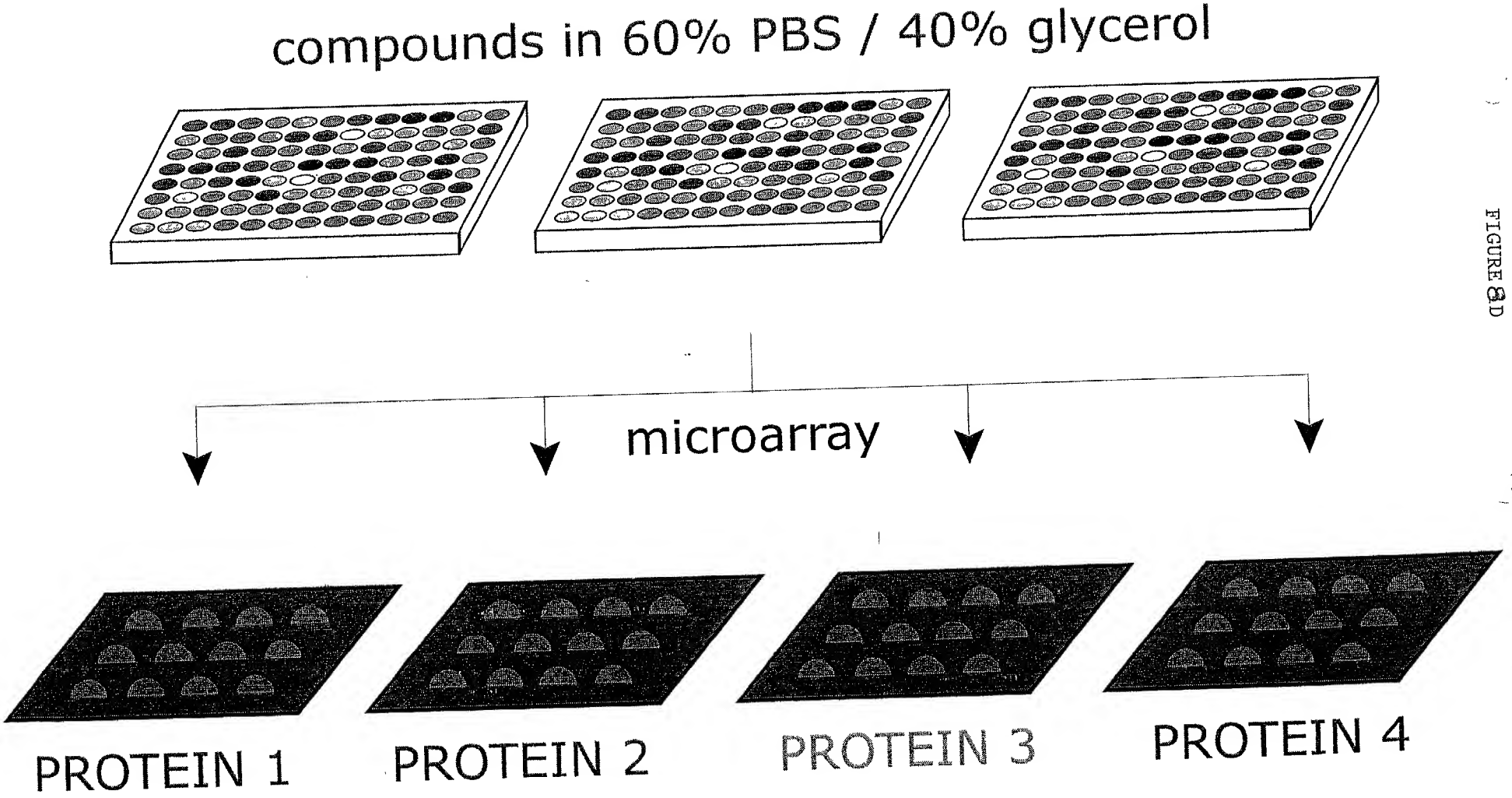


FIGURE 3D

TOE080 "EP2E2660

SCREENING METHOD 1

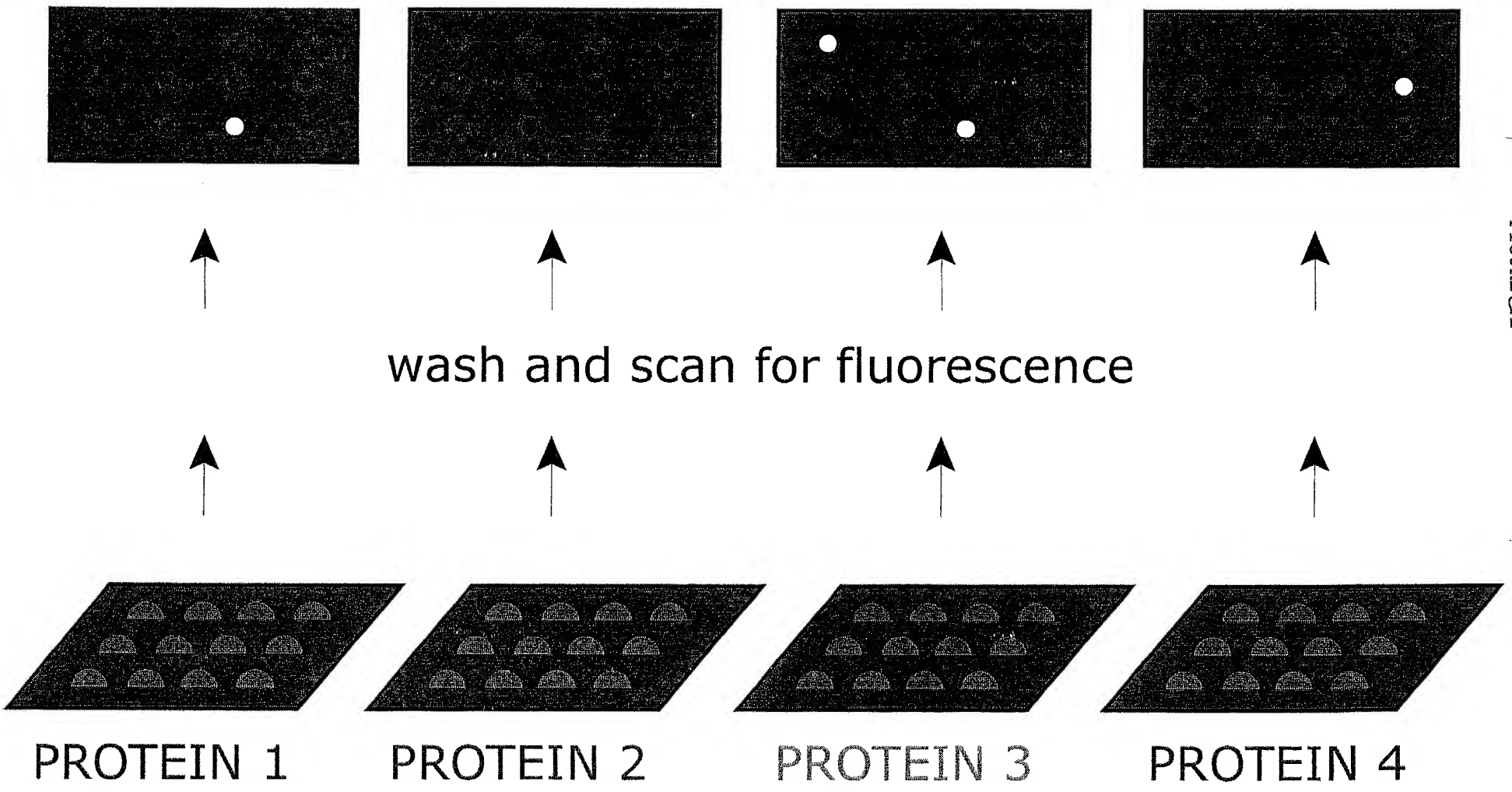


FIGURE 8E

TOC090 "Eh2E2660

SCREENING METHOD 1

On slide

"5-helix"
(a domain of gp120)

Peptide ligands

C37: 100 pM
DCC1: 500 pM
DCC2: 4000 nM

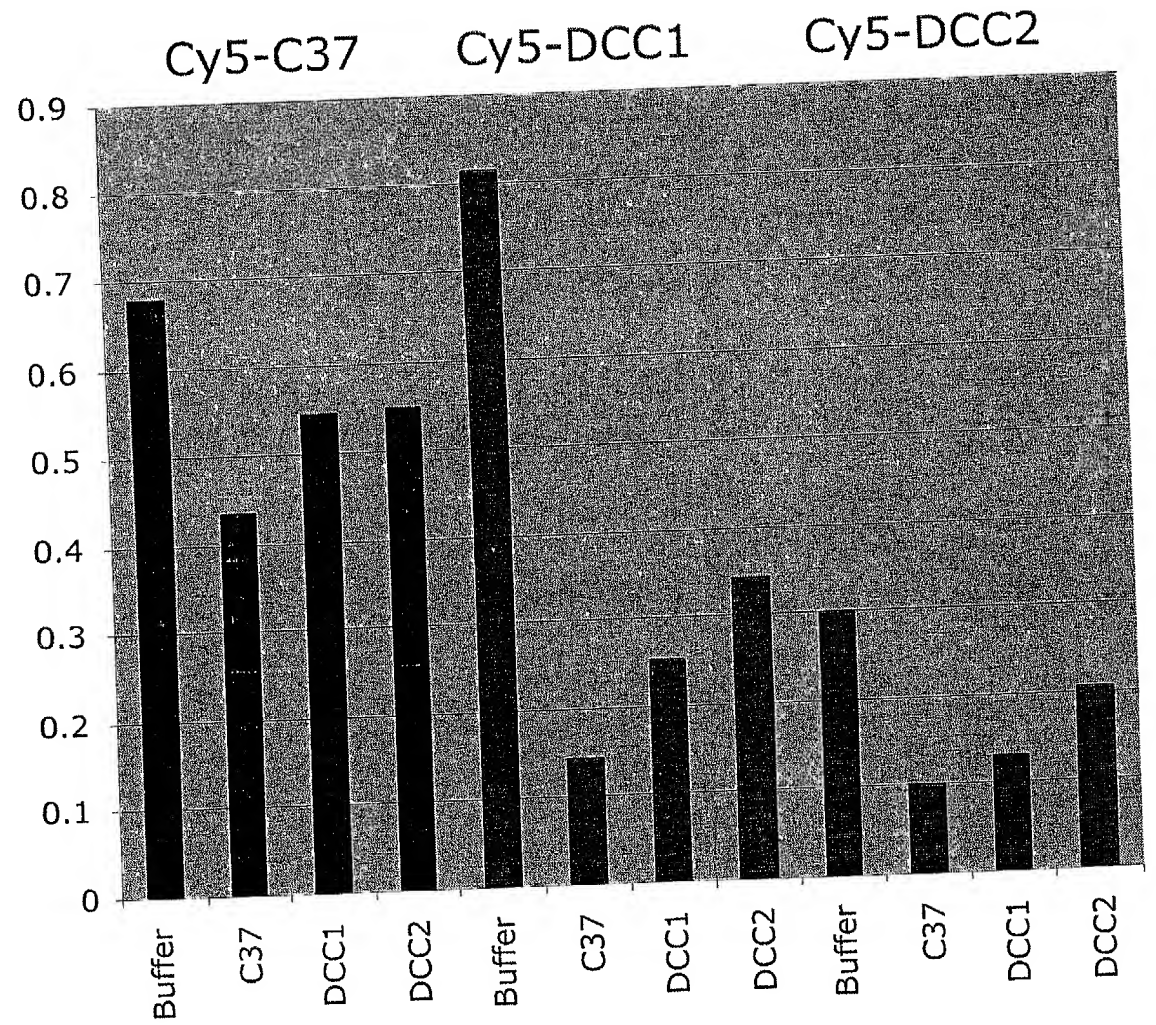


FIGURE 2